REMARKS/ARGUMENTS

The amendments set out above and the following remarks are responsive to the points raised by the Office Action dated February 21, 2008, and discussed during the interview with Examiner Sims on May 20, 2008. In view of the amendments set out above and the following remarks, reconsideration is respectfully requested.

As an initial point, Applicants' representative greatly appreciates the courtesy shown her by Examiner Sims, and further appreciates his careful consideration of the arguments presented during the interview.

Allowable Subject Matter

The Applicants are pleased to note that the Office Action indicates that claims 109-111 would be allowable if rewritten in independent form to include the limitations of the base claim and any intervening claims.

The Pending Claims

Claims 9, 12, 15-18, 20, 21, 23, 26-28, 85, 87, 90-99, and 103-117 are pending. Claims 9, 21, 85, and 103-111 have been amended, and claims 115-117 have been added, to describe the invention more clearly. No new matter has been added, and support for the amended claim language may be found within the original specification, claims, and drawings.

Claims 9, 21, 85, and 109-111 are supported at, for example, paragraphs [0088]-[0090] of the specification and by Figure 10. Claims 115-117 are supported at, for example, paragraphs [0097]-[0098], [0104], and [0108].

Claim Objections

Claims 106-108 were objected to under 37 C.F.R. § 1.175(c), as being of improper dependent form for failing to further limit the subject matter of a previous claim. It is respectfully asserted that claims 106-108 are proper dependent claims.

As an initial point, dependent claims 106-108 do further limit the methods of the respective independent claims from which they depend. The transitional phrase "consisting of" excludes any method steps beyond the steps that are claimed. Thus, the dependent claims 106-108 add a further, *negative* limitation not present in the independent claims, i.e., the exclusion of any further additional steps.

The test as to whether a claim is a proper dependent claim is that it includes every limitation of the claim from which it depends (35 U.S.C. § 112, fourth paragraph) or that it could not conceivably be infringed by anything which would not also infringe the basic claim (MPEP § 608.01(n)).

Dependent claims 106-108 are proper dependent claims under each of these analyses. First, dependent claims 106-108 include all of the limitations of claims 9, 21, and 85, respectively, from which they depend. Specifically, claims 106-108 recite that the method "consists of" steps (a), (b), (c), (d), and (e) of the respective independent claim from which claims 106-108 depend. Each of the steps recited in the independent claims 9, 21, and 85 is, therefore, included in dependent claims 106-108, respectively.

Dependent claims 106-108 also could not conceivably be infringed by anything that would not also infringe the respective independent claim. If all of the steps of dependent claims 106-108, i.e., steps (a), (b), (c), (d), and (e), are performed, the respective independent claim is also infringed, whether any additional steps are performed or not.

Thus, for the reasons set forth above, it is respectfully submitted that claims 106-108 are proper dependent claims. Accordingly, the claim objection under Rule 1.175(c) is improper and should be withdrawn.

Claim Rejections under 35 U.S.C. § 112

Claims 103-105 were rejected under 35 U.S.C. § 112, second paragraph, as indefinite for failing to particularly point out and distinctly claim the subject matter which the applicant regards as the invention.

According to the Office Action, "the only emission readings of the double stranded DNA intercalating dye," is vague and indefinite because it is unclear if the wording is

supposed to indicate some type of filtration wherein the emission readings do not comprise any background noise and are indeed strictly the only emission reading coming from the intercalating dye itself or that there are only two emission readings performed during each thermal cycle.

The claims have been amended to improve the form of the claims and to more distinctly claim the subject matter which the Applicants regard as the invention. Thus, it is respectfully submitted that with these amendments to the claims, the rejections under 35 U.S.C. § 112, second paragraph have now been overcome and should be withdrawn.

Rejections under 35 U.S.C. § 103

Claims 9, 12, 15-18, 20-21, 23, 26-28, 85, 87, 90-99, and 100-105 were rejected under 35 U.S.C. § 103 as unpatentable over U.S. Patent No. 6,472,156 to Wittwer et al. (hereinafter, "Wittwer") in view of U.S. Patent Publication No. 2003/0186259 to Loeffler et al. (hereinafter, "Loeffler").

This rejection is respectfully traversed.

Independent claims 9, 21, and 85 have been amended to include the limitations of formerly pending claims 100-102, and to recite using the first emission amount and corresponding thermal cycle number to obtain a first emission vs. cycle curve and using the second emission amount and corresponding thermal cycle number to obtain a second emission vs. cycle curve. Claims 9, 21, and 85 have also been amended to recite obtaining a standard emission vs. cycle curve and comparing the first emission vs. cycle curve with the standard emission vs. cycle curve to obtain the amount of the first amplicon and comparing the second emission vs. cycle curve with the standard emission vs. cycle curve to obtain the amount of the second amplicon.

The Office Action correctly acknowledges that Wittwer does not specifically teach that the first and second amplicons have melting curves which do not overlap. According to the Office Action, Wittwer teaches analyzing multiple sequence samples simultaneously and gives examples of different melting curves for three samples. The Office Action alleges that it would have been obvious to one of ordinary skill in the art to apply the method taught by Wittwer to samples that do not overlap in their melting curves because analyzing samples or

amplicons without overlapping melting curves would have been a simple substitution of components.

The Applicants assert that it would *not* have been a simple substitution of components to apply the claimed method to quantify first and second amplicons that do not have overlapping melting curves, and the claimed methods would not have been obvious to one of ordinary skill in the art.

First, Wittwer does not teach or suggest the quantification of first and second amplicons, wherein the melting curves of the first and second amplicons do not overlap. While the Wittwer references does teach the *detection* of more than one loci, it is not possible for the Wittwer reference to *quantify* first and second amplicons simultaneously, as claimed in claims 9, 21, and 85. *Quantification* of first and second amplicons simultaneously is not possible unless the multiple amplicons to be quantified have melting curves that do not overlap, as claimed. If the melting curves overlap, it would not be possible to separately quantify each amplicon because the first amplicon will be giving a first emission reading at the same time that the second amplicon is giving a second emission reading. With first and second emission readings being detected at the same time, it would not be possible to separately *quantify* each amplicon without mixing up the signal of one amplicon with those of the other amplicon. In Wittwer, the overlapping melting curves of first and second loci would provide two different peaks, which would enable the *detection* of a first and second locus, however, the *quantification* of first and second amplicons would not be possible. Thus, the non-overlapping nature of the melting curves of the first and second amplicons of the claimed method makes it possible to quantify both amplicons, which Wittwer cannot do.

Secondly, in order to quantify the amplicons, emission readings must be obtained during *each thermal cycle* of the PCR protocol to obtain the first and second melting curves, which are in turn used to obtain the amounts of the first and second amplicons, as claimed in claims 9, 21, and 85. Wittwer, in contrast, does not obtain emission readings during each thermal cycle of the PCR. The method of Wittwer can go no further than the mere *detection* of multiple loci. Because Wittwer does not take emission readings during *each* thermal cycle, as claimed, Wittwer cannot obtain a melting curve, and it is not possible for the method of Wittwer to quantify first and second amplicons, as claimed.

Far from being a simple substitution of known components, the non-overlapping nature of the melting curves makes it possible to *quantify* (and not only detect) more than one amplicon in a single PCR reaction. Wittwer, in contrast, provides absolutely no guidance or teaching with respect to how to quantify more than one amplicon in a single PCR reaction, let alone any guidance with respect to non-overlapping melting curves or measuring emission amounts during each thermal cycle. Thus, the obviousness rejection cannot be maintained.

The Office Action alleges that Figure 1 shows different melting curves. Figure 1 of Wittwer, however, is not applicable to the claimed method. In Figure 1, Wittwer teaches genotyping three different samples using a probe that hybridizes to the locus being analyzed. The melting temperature of the probe depends on the percent identity of the probe to the target complementary sequence. The greater the difference between the probe and the target complementary sequence, the lower the temperature needed to separate the strands (Wittwer col. 12, lines 20-59). Figure 1 shows the most stable mismatch of the probe with the mutant sample (i.e., a G-T mismatch), which Wittwer explains results in a melting temperature shift of only 2-3° C. Therefore, Figure 1 of Wittwer merely shows that the mismatched hybridization of a probe to a sample produces a detectable melting curve shift between a wild-type and a mutant sample. Thus, Wittwer Figure 1, which genotypes three different samples, teaches nothing about non-overlapping melting curves of multiple amplicons to be quantified in a single PCR reaction, as claimed.

In fact, Figure 1 of Wittwer teaches away from even detecting first and second amplicons with melting curves that do not overlap. In Figure 1 of Wittwer, the melting curves for the hybridization of the probe with the homozygous wild type, the heterozygote, and the homozygous mutant are shown and clearly overlap substantially. As shown in Figure 1, Wittwer's genotyping and detection method involves a substantial amount of melting curve overlap between the samples, unlike the presently claimed method, which requires that the melting curves of the first and second amplicons to be quantified do not overlap. Accordingly, the obviousness rejection cannot be maintained.

The Office Action correctly acknowledges that Wittwer does not explicitly teach the limitation of quantifying the first and second amplicons. Wittwer, indeed, teaches nothing about how to *quantify* first and second amplicons and gives no guidance to one of ordinary

skill in the art as to how quantification can be accomplished. Wittwer's instructions are limited to *detecting* DNA sequences. Without giving any teaching or guidance as to how one of ordinary skill even goes about quantifying amplicons, Wittwer can provide no guidance whatsoever regarding whether the melting curves of the amplicons overlap. Accordingly, the obviousness rejection cannot stand.

Moreover, Wittwer fails to teach quantifying the first and second amplicons comprising, *inter alia*, using the first and second emission amounts and corresponding thermal cycle number to obtain first and second emission vs. cycle curves, and obtaining a standard emission vs. cycle curve and comparing the first and second emission vs. cycle curves with the standard emission vs. cycle curve to obtain the amount of the first and second amplicons. As explained above, Wittwer, indeed, teaches nothing about how to *quantify* first and second amplicons and gives no guidance to one of ordinary skill in the art as to how quantification can be accomplished. Wittwer's teachings with respect to detecting sequences falls short of teaching the claimed steps of obtaining first and second emission vs. cycle curves and comparing them to a standard emission vs. cycle curve.

Loeffler fails to cure the deficiencies of Wittwer. Loeffler fails to teach the quantification of first and second amplicons, wherein the melting curves of the first and second amplicon do not overlap. Loeffler also fails to teach using the first and second emission amounts and corresponding thermal cycle number to obtain first and second emission vs. cycle curves, and obtaining a standard emission vs. cycle curve and comparing the first and second emission vs. cycle curves with the standard emission vs. cycle curve to obtain the amount of the first and second amplicons, as claimed. Accordingly, the combination of Wittwer and Loeffler also fails to render the presently claimed invention obvious.

The Applicants also note that dependent claims 106-108 and 112-114 were not rejected over any prior art. The Applicants assert that dependent claims 106-108 and 112-114 are also patentable over the cited references.

Because the independent claims are patentable for the reasons set forth above, the dependent claims are also patentable because they depend from patentable independent claims.

Conclusion

Applicants respectfully submit that the patent application is in condition for allowance. If, in the opinion of the Examiner, a telephone conference would expedite the prosecution of the subject application, the Examiner is invited to call the undersigned attorney.

Respectfully submitted,

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